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(21) International Application Number: PCT/US93/09001 (22) International Filing Date: 23 September 1993 (23.09.93) (30) Priority data: 07/968,770 30 October 1992 (30.10.92) US (60) Parent Application or Grant (63) Related by Continuation US 07/968,770 (CIP) Filed on 30 October 1992 (30.10.92) (71) Applicant (for all designated States except US): MERRELL DOW PHARMACEUTICALS INC. [US/US]; 2110 East Galbraith Road, P.O. Box 156300, Cincinnati, OH 45215-6300 (US).	(72) Inventor; and (75) Inventor/Applicant (for US only) : FLYNN, Gary, A. [US, US]; 7121 Euclid Road, Cincinnati, OH 45243 (US). (74) Agent: BARNEY, Charlotte, L.; Marion Merrell Dow Inc., 2110 East Galbraith Road, P.O. Box 156300, Cin cinnati, OH 45215-6300 (US). (81) Designated States: AT, AU, BB, BG, BR, BY, CA, CH CZ, DE, DK, ES, FI, GB, HU, JP, KP, KR, KZ, LK LU, MG, MN, MW, NL, NO, NZ, PL, PT, RO, RU SD, SE, SK, UA, US, VN, European patent (AT, BE CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA GN, ML, MR, NE, SN, TD, TG). Published <i>With international search report.</i>	
(54) Title: NOVEL MERCAPTOACETYLAMIDE BICYCLIC LACTAM DERIVATIVES USEFUL AS INHIBITORS OF ENKEPHALINASE AND ACE (57) Abstract The present invention relates to certain novel mercaptoacetylamine bicyclic lactam derivatives useful as inhibitors of enkephalinase and of ACE.		

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(54) Title: NOVEL MERCAPTOACETYLAMIDE BICYCLIC LACTAM DERIVATIVES USEFUL AS INHIBITORS OF ENKEPHALINASE AND ACE (57) Abstract The present invention relates to certain novel mercaptoacetylamine bicyclic lactam derivatives useful as inhibitors of enke- phalinase and of ACE.		

5

10 NOVEL MERCAPTOACETYLAMIDE BICYCLIC LACTAM DERIVATIVES
 USEFUL AS INHIBITORS OF ENKEPHALINASE AND ACE

BACKGROUND OF THE INVENTION

15 This is a continuation-in-part of application Serial No.
 07/968,770, filed October 30, 1992.

 Enkephalinase or, more specifically, endopeptidase-
24.11, is a mammalian ectoenzyme which is involved in the
20 metabolic degradation of certain circulating regulatory
 peptides. This enzyme, which is a Zn^{+2} -metallopeptidase,
 exerts its effect by cleaving the extracellular peptides at
 the amino group of hydrophobic residues and thus
 inactivates the peptides as regulatory messengers.

25

 Enkephalinase is involved in the metabolic degradation
 of a variety of circulating regulatory peptides including
 endorphins, such as β -endorphin and the enkephalins, atrial
 natriuretic peptide (ANP), and other circulating regulatory
30 peptides.

 Endorphins are naturally-occurring polypeptides which
 bind to opiate receptors in various areas of the brain and
 thereby provide an analgesic effect by raising the pain
35 threshold. Endorphins occur in various forms including α -
 endorphin, β -endorphin, γ -endorphin as well as the
 enkephalins. The enkephalins, i.e., Met-enkephalin and Leu-
 enkephalin, are pentapeptides which occur in nerve endings

of brain tissue, spinal cord and the gastrointestinal tract. Like the other endorphins, the enkephalins provide an analgesic effect by binding to the opiate receptors in the
5 brain. By inhibiting enkephalinase, the metabolic degradation of the naturally-occurring endorphins and enkephalins are inhibited, thereby providing a potent endorphin- or enkephalin-mediated analgesic effect. Inhibition of enkephalinase would therefore be useful in a
10 patient suffering from acute or chronic pain. Inhibition of enkephalinase would also be useful in providing an antidepressant effect and in providing a reduction in severity of withdrawal symptoms associated with termination of opiate or morphine administration. In addition,
15 inhibition of enkephalinase would also be useful in the treatment of irritable bowel syndrome

ANP refers to a family of naturally-occurring peptides which are involved in the homeostatic regulation of blood
20 pressure, as well as sodium and water levels. ANP have been found to vary in length from about 21 to about 126 amino acids with a common structural feature being one or more disulfide-looped sequences of 17 amino acids with various amino- and carboxy-terminal sequences attached to the
25 cystine moiety. ANP have been found to bind to specific binding sites in various tissues including kidney, adrenal, aorta, and vascular smooth muscle with affinities ranging from about 50 pico-molar (pM) to about 500 nano-molar (nM) [Needleman, *Hypertension* 7, 469 (1985)]. In addition, it is
30 believed that ANP binds to specific receptors in the brain and possibly serves as a neuromodulator as well as a conventional peripheral hormone.

The biological properties of ANP involve potent
35 diuretic/natriuretic and vasodilatory/hypotensive effects as well as an inhibitory effect on renin and aldosterone secretion [deBold, *Science* 230, 767 (1985)]. By inhibiting enkephalinase, the metabolic degradation of the naturally-

-3-

occurring ANP are inhibited, thereby providing a potent ANP-mediated diuretic, natriuretic, hypotensive, hypoaldosteronemic effects. Inhibition of enkephalinase
5 would therefore be useful in a patient suffering from disease states characterized by abnormalities in fluid, electrolyte, blood pressure, intraocular pressure, renin, or aldosterone homeostasis, such as, but not limited to, hypertension, renal diseases, hyperaldosteronemia, cardiac
10 hypertrophy, glaucoma and congestive heart failure.

In addition, the compounds of the present invention are inhibitors of Angiotension-Converting Enzyme (ACE). ACE is a peptidyl dipeptidase which catalyzes the conversion of
15 angiotensin I to angiotensin II. Angiotensin II is a vasoconstrictor which also stimulates aldosterone secretion by the adrenal cortex. Inhibition of ACE would therefore be useful in a patient suffering from disease states such as hypertension and congestive heart failure [See William W.
20 Douglas, "Polypeptides - Angiotensin, Plasma Kinins, and Others", Chapter 27, in GOODMAN AND GILLMAN'S THE PHARMACOLOGICAL BASIS OF THERAPEUTICS, 7th edition, 1985, pp. 652-3, MacMillan Publishing Co., New York, New York]. In addition, it has been discovered that ACE inhibitors are
25 useful in treating cognitive disorders [German Application No. 3901-291-A, published August 3, 1989].

Bradykinin refers to a naturally-occurring peptide which is a very powerful vasodilator and causes increased
30 capillary permeability. By inhibiting enkephalinase and ACE, the metabolic degradation of bradykinin is inhibited, thereby providing increased levels of bradykinin in the circulation.

35 In addition, the compounds of the present invention are useful as inhibitors of smooth cell proliferation. Smooth muscle cell proliferation in the intima of muscular arteries is a primary cause of vascular stenosis in arteriosclerosis,

after vascular surgery, and after coronary angioplasty. Several animal studies have indicated the renin-angiotensin system plays an important role in this vascular response to injury. Chronic treatment with angiotensin converting enzyme (ACE) inhibitors reduced myointimal thickening following balloon injury in rat carotid artery or aorta. Powell, J.S., Muller, R.K.M. and Baumgartner, H.R.; Suppression of the vascular response to injury: The role of angiotensin-converting enzyme inhibitors. J. Am. Coll. Cardiol. 17:137B-42B, 1991. More recently, atrial natriuretic peptide (ANP) has been found to decrease myointimal proliferation. ANP is rapidly metabolized by receptor mediated clearance and by neutral endopeptidase (NEP). Inhibition of NEP significantly reduces proliferation in the balloon-injured rabbit vasculature. Davis, H.R., McGregor, D.C., Hoos, L., Mullins, D.E. and Sybertz, E.J.: Atrial natriuretic factor and the neutral endopeptidase inhibitor SCH42495 prevent myointimal proliferation after vascular injury. Circ. 86:I-220, 1992. These studies imply that a dual inhibitor of ACE and NEP should be therapeutically useful in the treatment of conditions which require inhibition of smooth cell proliferation. Davis and Sybertz, European Patent Application 533084-A1, March 24, 1993.

SUMMARY OF THE INVENTION

The present invention provides novel compounds of the Formula (I) wherein

R is hydrogen, a C₁-C₄ alkyl or an Ar-Y- group, -CH₂O-C(O)C(CH₃)₃ or diphenylmethyl;

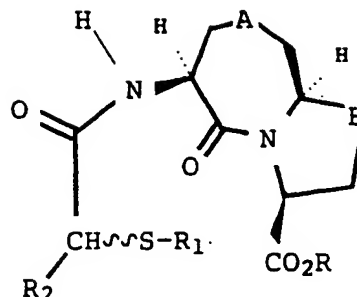
R₁ is hydrogen, acetyl, -CH₂O-C(O)C(CH₃)₃ or benzoyl or a group of the formula

R₂ is hydrogen, C₁-C₈ alkyl, -CH₂OCH₂CH₂OCH₃ or an Ar-Y-group;

A is -CH₂-, -O-, or -S-;

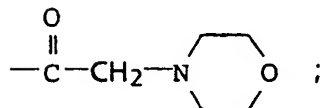
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(I)

15



20

B is -S- or -O-; and
and the pharmaceutically acceptable salts thereof.

25

The present invention further provides a method of inhibiting enkephalinase in a patient in need thereof comprising administering to said patient an effective
enkephalinase inhibitory amount of a compound of Formula (I). The present invention also provides a method of inhibiting ACE in a patient in need thereof comprising administering to said patient an effective ACE inhibitory amount of a compound of Formula (I).

30

35

In addition, the present invention provides a composition comprising an assayable amount of a compound of Formula (I) in admixture or otherwise in association with an inert carrier. The present invention also provides a
pharmaceutical composition comprising an effective inhibitory amount of a compound of Formula (I) in admixture or otherwise in association with one or more pharmaceutically acceptable carriers or excipients.

DETAILED DESCRIPTION OF THE INVENTION

5 As used herein, the term "C₁-C₄ alkyl" refers to a saturated straight or branched chain hydrocarbyl radical of one to four carbon atoms and includes methyl, ethyl, propyl, isopropyl, n-butyl, isobutyl, tertiary butyl and the like. The term "C₁-C₄ alkoxy" refers to a saturated straight or
10 branched chain hydrocarboxy radical of one to four carbon atoms and includes methoxy, ethoxy, propoxy, isopropoxy, n-butoxy, isobutoxy, tertiary butoxy and the like.

As used herein, the term "Ar-Y-" refers to a radical
15 wherein Ar is an aryl group and Y is a C₀-C₄ alkyl. The term "Ar" refers to a phenyl, 2-benzofuranyl or naphthyl group unsubstituted or substituted with from one to three substituents selected from the group consisting of methylenedioxy, hydroxy, C₁-C₄ alkoxy, fluoro and chloro.
20 The term "C₀-C₄ alkyl" refers to a saturated straight or branched chain hydrocarbyl radical of zero to four carbon atoms and includes a bond, methyl, ethyl, propyl, isopropyl, n-butyl, isobutyl, tertiary butyl and the like. Specifically included within the scope of the term "Ar-Y-"
25 are phenyl, naphthyl, phenylmethyl or benzyl, phenylethyl, p-methoxybenzyl, 3,4-methylenedioxy, p-fluorobenzyl and p-chlorobenzyl.

As used herein, the designation "∩" refers to a bond to
30 a chiral atom for which the stereochemistry is not designated.

Compounds of Formula (I) can form pharmaceutically acceptable salts with any non-toxic, organic or inorganic
35 acid. Illustrative inorganic acids which form suitable salts include hydrochloric, hydrobromic, sulphuric and phosphoric acid and acid metals salts such as sodium monohydrogen orthophosphate and potassium hydrogen sulfate.

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Illustrative organic acids which form suitable salts include the mono, di and tricarboxylic acids. Illustrative of such acids are, for example, acetic, trifluoroacetic, glycolic, 5 lactic, pyruvic, malonic, succinic, glutaric, fumaric, malic, tartaric, citric, ascorbic, maleic, hydroxymaleic, benzoic, hydroxybenzoic, phenylacetic, cinnamic, salicylic, 2-phenoxybenzoic and sulfonic acids such as methane sulfonic, trifluoromethane sulfonic, 2-hydroxyethane 10 sulfonic acid and p-toluenesulfonic acid.

The compounds of Formula (I) can be prepared by utilizing procedures and techniques well known and appreciated by one of ordinary skill in the art. A general 15 synthetic scheme for preparing these compounds is set forth in Scheme A wherein all substituents, unless other indicated, are as previously defined.

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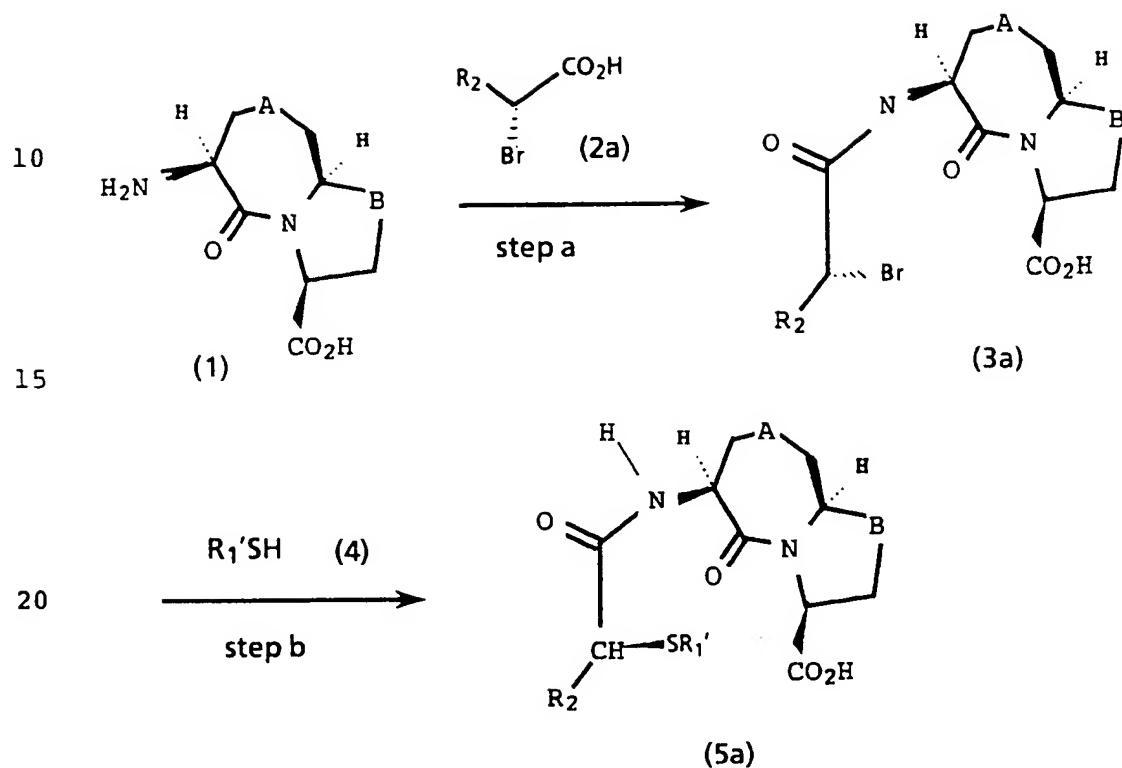
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Scheme A

5



25 $R_1' = \text{COCH}_3 \text{ or } \text{COPh}$

In step a, the appropriate bicyclic lactam compound of structure (1) is reacted with the appropriate (S)-bromoacid of structure (2a) to give the corresponding (S)-bromoamide compound of structure (3a). For example, the appropriate bicyclic lactam compound of structure (1) can be reacted with the appropriate (S)-bromoacid of structure (2a) in the presence of a coupling reagent such as EEDQ (1-ethoxycarbonyl-2-ethoxy-1,2-dihydroquinoline), DCC (1,3-dicyclohexylcarbodiimide), or diethylcyanophosphonate in a suitable aprotic solvent, such as methylene chloride to give the appropriate (S)-bromoamide compound of structure (3a).

Alternatively the appropriate bicyclic lactam compound of structure (1) is reacted with the appropriate (R)-bromoacid to give the corresponding (R)-bromoamide or the
5 appropriate bicyclic lactam compound of structure (1) is reacted with the appropriate enantiomeric mixture of the bromoacid to give the corresponding enantiomeric mixture of bromoamide as described.

10 In step b, the (S)-bromo functionality of the appropriate (S)-bromoamide compound of structure (3a) is converted to the corresponding (R)-thioacetate or (R)-thiobenzoate of structure (5a).

15 For example, the appropriate (S)-bromoamide compound of structure (3a) is reacted with thiolacetic acid or thiolbenzoic acid of structure (4) in the presence of a base, such as cesium carbonate. The reactants are typically contacted in a suitable organic solvent such as a mixture of
20 dimethylformamide and tetrahydrofuran. The reactants are typically stirred together at room temperature for a period of time ranging from 1 to 8 hours. The resulting (R)-thioacetate or (R)-thiobenzoate of structure (5a) is recovered from the reaction zone by extractive methods as is
25 known in the art. It may be purified by chromatography.

Alternatively, the (R)-bromo functionality of the appropriate (R)-bromoamide is converted to the corresponding (S)-thioacetate or (S)-thiobenzoate or the bromo
30 functionality of the appropriate enantiomeric mixture of of the bromoamide wherein is converted to the corresponding enantiomeric mixture of thioacetate or thiobenzoate compounds.

35 As summarized in Table 1, the R and R₁ groups on the compounds of structures (5) can be manipulated using techniques and procedures well known and appreciated by one

of ordinary skill in the art to give the corresponding compounds of structures (6)-(12).

5 The (R)-thioacetate or (R)-thiobenzoate functionality of the appropriate compound of structure (5) can be removed with lithium hydroxide in a suitable solvent mixture such as tetrahydrofuran and ethanol to give the appropriate (R)-thio compound of structure (6).

10

 Alternatively, the carboxylic acid functionality of the appropriate compound of structure (5) can be re-esterified using techniques and procedures well known and appreciated in the art. For example, a compound of structure (7) can be
15 prepared by treating the carboxylic acid compound of structure (5) with the appropriate alkyl halide in a suitable aprotic solvent, such as dimethylformamide along with a non-nucleophilic base, such as cesium carbonate.

20 The (R)-thioacetate or (R)-thiobenzoate functionalities of the appropriate compounds of structure (7) can be hydrolyzed to the corresponding (R)-thiol compounds of structure (8) with ammonia in a suitable protic solvent, such as methanol.

25

 The thiol functionality of the appropriate compound of structure (6) can be alkylated using techniques and procedures well known and appreciated in the art. For example, a compound of structure (9) can be prepared by
30 treating the thiol compound of structure (6) with chloromethyl pivalate in a suitable aprotic solvent, such as dimethylformamide along with a non-nucleophilic base, such as cesium carbonate or pyridine.

35 The thiol functionality of the appropriate compound of structure (8) can be alkylated using techniques and procedures well known and appreciated in the art. For example, a compound of structure (10) can be prepared by

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treating the thiol compound of structure (8) with chloromethyl pivalate as described above for the conversion of (6) to (9).

5

The thiol functionality of the appropriate compound of structure (6) can be acylated to give the 4-morpholinoacetyl compound of structure (11). For example, a compound of structure (11) can be prepared by treating the thiol
10 compound of structure (6) with 4-morpholinethiolacetate in the presence of a coupling reagent such as DCC in a suitable aprotic solvent such as methylene chloride.

In addition, a 4-morpholinoacetyl compound of structure
15 (11) can be prepared by treating the (R) or (S)-bromoamide compound of structure (3) wherein with triphenylmethyl 4-morpholinethiolacetate in the presence of a base, such as sodium hydride, in a suitable aprotic solvent such as dimethylformamide.

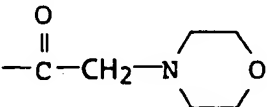
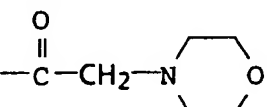
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The thiol functionality of the appropriate compound of structure (8) can acylated to give the 4-morpholinoacetyl compound of structure (12). For example, a compound of structure (12) can be prepared by treating the thiol
25 compound of structure (8) with 4-morpholinethiolacetate in the presence of a coupling reagent such as DCC in a suitable aprotic solvent such as methylene chloride.

30

35

TABLE 1
MANIPULATION OF R AND R₁

5	Compound	R	R ₁
	5	H	COCH ₃ or COPh
	6	H	H
10	7	C ₁ -C ₄ alkyl, Ar-Y, -CH ₂ OCOC(CH ₃) ₃ , diphenylmethyl	COCH ₃ or COPh
	8	C ₁ -C ₄ alkyl, Ar-Y, -CH ₂ OCOC(CH ₃) ₃ , diphenylmethyl	H
15	9	H	-CH ₂ OCOC(CH ₃) ₃
	10	C ₁ -C ₄ alkyl, Ar-Y, -CH ₂ OCOC(CH ₃) ₃ , diphenylmethyl	-CH ₂ OCOC(CH ₃) ₃
20	11	H	
25	12	C ₁ -C ₄ alkyl, Ar-Y, -CH ₂ OCOC(CH ₃) ₃ , diphenylmethyl	

Starting materials for use in the general synthetic procedures outlined in Scheme A are readily available to one of ordinary skill in the art. For example, [3R-(3 α ,6 α ,9 α)-
30 6-aminooctahydro-5-oxo-thiazolo[3,2-a]azepine-3-carboxylic acid may be prepared as described in United States Patent 4,415,496 (November 15, 1983).

Alternatively, the bicyclic lactam starting materials of
35 structure (1) may be prepared as set forth in Scheme B. In Scheme B, all substituents are as previously described unless otherwise indicated.

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Scheme B

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10

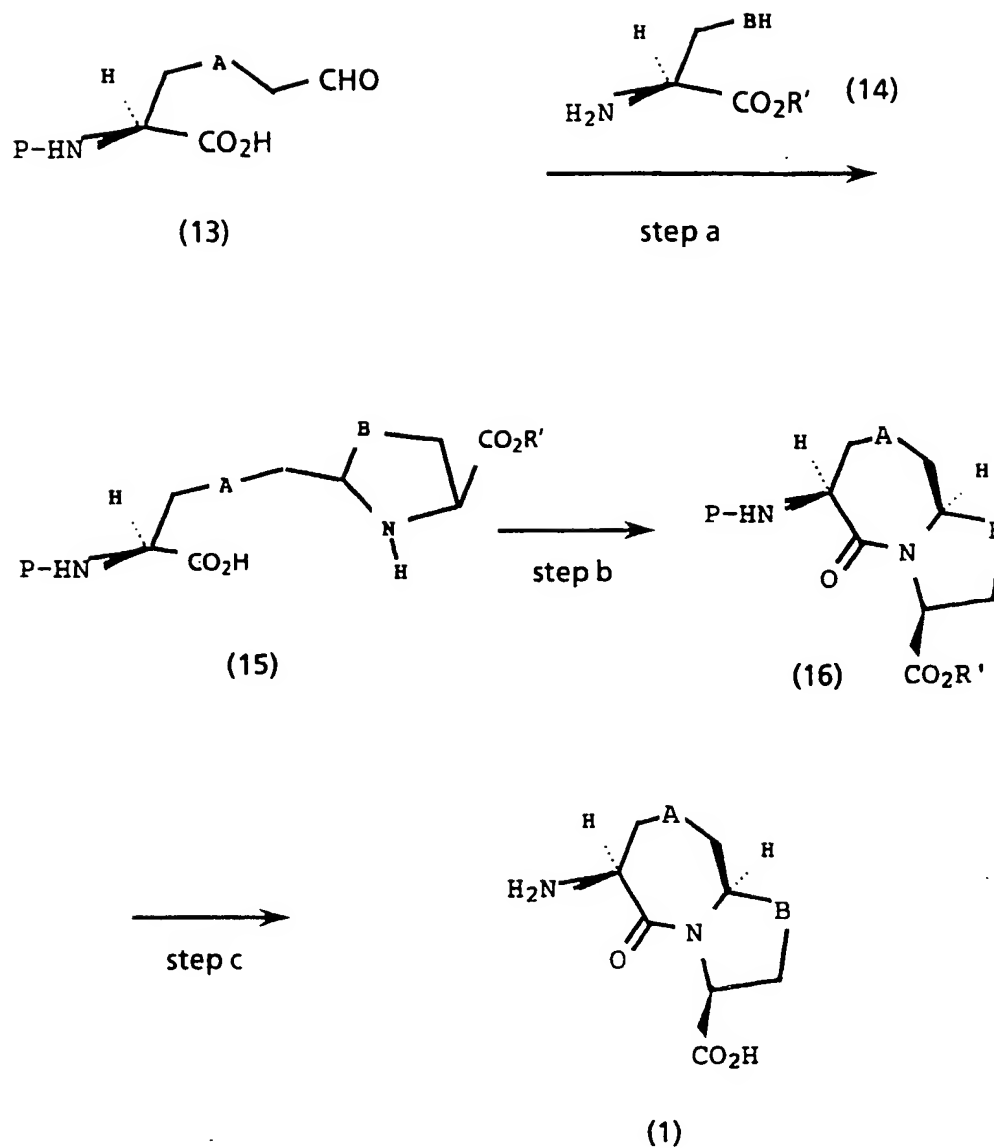
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P = Boc, CBZ or Phth
R' = Me or Et

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In step a, the aldehyde compound of structure (13) is condensed with an ester of L-serine or L-cysteine (14) to give a diastereomeric mixture of oxazolidines or thiazolidines of structure (15).

In step b, the diastereomeric mixture of oxazolidines or thiazolidines of structure (15) is cyclized with N-ethoxycarbonyl-2-ethoxy-1,2-dihydroquinoline to form the protected (S,R,R) and (S,S,R) bicyclic lactams. The protected bicyclic lactams are then separated to give the protected bicyclic lactam of structure (16).

In step c, the protected bicyclic lactam of structure (16) is deprotected to give the bicyclic lactam of structure (1).

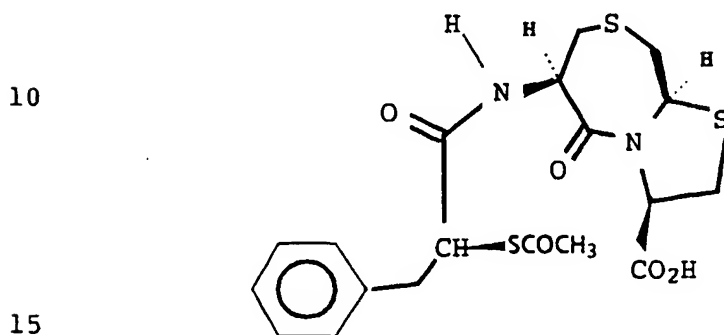
Starting materials for use in Scheme B are readily available to one of ordinary skill in the art. For example, 5-formyl-2(S)-phthalimidopentanoic acid is described in United States Patent 4,415,496 (November 15, 1983).

The following examples present typical syntheses as described in Scheme A. These examples are understood to be illustrative only and are not intended to limit the scope of the present invention in any way. As used herein, the following terms have the indicated meanings: "g" refers to grams; "mmol" refers to millimoles; "mL" refers to milliliters; "bp" refers to boiling point; "°C" refers to degrees Celsius; "mm Hg" refers to millimeters of mercury; "μL" refers to microliters; "μg" refers to micrograms; and "μM" refers to micromolar.

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Example 1

[3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]thiazepine-3-carboxylic acid



Scheme B, step a: Ethyl 2-[[[2'-carboxy-2'-benzoyloxycarbonyl]ethyl]ethylsulfide]-4(R)-thiazolidinecarboxylate

20 Wash sodium hydride (7.75g, 191mmol of a 59% dispersion in paraffin) 2 times with dry hexane (2X) under a nitrogen atmosphere. Add anhydrous dimethylformamide (90mL) and cool with an ice/methanol bath. A, by portionwise addition, L-cysteine ethyl ester hydrochloride (96.7mmol), stir for 5

25 minutes and add potassium iodide (5.2g, 32mmol). Add, by dropwise addition, bromoacetaldehyde diethylacetal (14.5mL, 96.7mmol), remove the ice bath and stir for 8 hours at room temperature. Evaporate the solvent *in vacuo* to give S-(2-Diethoxyethyl)-L-cysteine ethyl ester which is used in the

30 next step without purification.

Mix S-(2-Diethoxyethyl)-L-cysteine ethyl ester (6.6mmol) and pyridine (60mL). Add, by dropwise addition, benzyl chloroformate (7.3mmol) and stir overnight. Remove excess

35 pyridine in vacuo and dissolve the residue in a two-phase mixture of ethyl acetate/water. Separate the organic phase and extract the aqueous phase with additional ethyl acetate (2X). Wash the combined organic phases with water, then

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brine and dry (MgSO₄). Evaporate the solvent *in vacuo* to give N-(benzyloxycarbonyl)-S-(2-diethoxyethyl)-L-cysteine ethyl ester.

5

Dissolve N-(benzyloxycarbonyl)-S-(2-diethoxyethyl)-L-cysteine ethyl ester (21.7mmol) in ethanol (150mL. Add 1N lithium hydroxide (50mL) and stir overnight at room temperature. Carefully adjust to pH 4 with 1N hydrochloric
10 acid and stir for 1 hour. Extract into ethyl acetate, dry (MgSO₄) and evaporate the solvent *in vacuo* to give N-(benzyloxycarbonyl)-S-(formylmethyl)-L-cysteine.

Dissolve L-cysteine ethyl ester (12.7mmol) in tetrahydrofuran
15 (120mL) and add N-(benzyloxycarbonyl)-S-(formylmethyl)-L-cysteine (12.7mmol). Place under a nitrogen atmosphere and stir for 3 hours. Evaporate the solvent *in vacuo*, dissolve the residue in chloroform and wash with water (2X30mL). Combine the aqueous extracts and extract with chloroform
20 (2X30mL). Combine all organic extracts, dry (Na₂SO₄) and evaporate the solvent *in vacuo* to give the title compound.

Scheme B, step b: Ethyl [3R-[3 α ,6 α , (S*),9 α]]-6-
[benzyloxycarbonylamino]octahydro-5-oxothiazolo[3,2-
25 a][1,4]thiazepine-3-carboxylate and Ethyl [3R-
[3 α ,6 α , (S*),9 α]]-6-[benzyloxycarbonylamino]octahydro-5-
oxothiazolo[3,2-a][1,4]thiazepine-3-carboxylate

Dissolve ethyl 2-[[[2'-carboxy-2'-benzyloxycarbonyl]ethyl]ethylsulfide]-4(R)-
30 thiazolidinecarboxylate (12.7mmol) in tetrahydrofuran (30mL) and add N-ethoxycarbonyl-2-ethoxy-1,2-dihydroquinoline (EEDQ) (469mg). Stir overnight at room temperature under a nitrogen atmosphere. Evaporate the solvent *in vacuo*, partition between ethyl acetate and dilute hydrochloric
35 acid. Separate the organic phase, wash with 5% sodium bicarbonate solution, water and brine. Dry (Na₂SO₄), evaporate the solvent *in vacuo* and purify and separate the

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isomers by silica gel chromatography to give the separate isomeric title compounds.

5 Scheme B, step c: [3R-[3 α ,6 α , (S*),9 α]]-6-amino-octahydro-5-oxothiazolo[3,2-a][1,4]thiazepine-3-carboxylic acid

- Dissolve ethyl [3R-[3 α ,6 α , (S*),9 α]]-6-[benzyloxycarbonylamino]octahydro-5-oxothiazolo[3,2-a][1,4]thiazepine-3-carboxylate (21.7mmol) in ethanol
10 (150mL). Add 1N lithium hydroxide (50mL) and stir overnight at room temperature. Concentrate *in vacuo*. Partition between ethyl acetate and 6N hydrochloric acid. Separate the organic phase and wash with brine. Dry (MgSO₄) and evaporate the solvent *in vacuo* to give [3R-[3 α ,6 α , (S*),9 α]]-
15 6-[benzyloxycarbonylamino]octahydro-5-oxothiazolo[3,2-a][1,4]thiazepine-3-carboxylic acid.

- Mix [3R-[3 α ,6 α , (S*),9 α]]-6-[benzyloxycarbonylamino]octahydro-5-oxothiazolo[3,2-
20 a][1,4]thiazepine-3-carboxylic acid (27.7mmol), trifluoroacetic acid (75mL) and anisole (5mL). Stir at room temperature overnight. Pour onto water and carefully neutralize with solid sodium hydrogen carbonate. Extract into ethyl acetate (2X), wash with brine and dry (MgSO₄).
25 Evaporate the solvent *in vacuo* and purify by chromatography to give the title compound.

Scheme A, step a: [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(R)-bromo-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-
30 a][1,4]thiazepine-3-carboxylic acid

- Mix D-phenylalanine (186.4g, 1.128mol) and 49% hydrobromic acid (372.8g), cool to -5°C and add, by dropwise addition, a solution of sodium nitrite (77.9g) in water (565mL) over a period of 1 hour (vigorous gas evolution). Stir at -5°C to
35 0°C for 4 hours, extract into ethyl ether (3X1L), dry (MgSO₄) and evaporate the solvent *in vacuo*. Purify by chromatography (5% acetic acid/95% methylene chloride) and

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distill to give 3-phenyl-2(R)-bromopropionic acid (112g, 43%); bp 128-135°C @ 0.25 torr.

5 Mix 3-phenyl-2(R)-bromopropionic acid (1.0mmol) and [3R-[3 α ,6 α , (S*),9 α]]-6-amino-octahydro-5-oxothiazolo[3,2-a][1,4]thiazepine-3-carboxylic acid (1.0mmol) in methylene chloride (6mL). Add EEDQ (247mg, 1.0mmol). Stir for 15 hours at ambient temperature under argon atmosphere. Dilute
10 with ethyl acetate (25mL) and wash with 5% sulfuric acid (15mL), then saturated sodium hydrogen carbonate (15mL). Dry (Na₂SO₄), concentrate *in vacuo* and purify by silica gel chromatography to yield the title compound.

15 Scheme A, step b: [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]thiazepine-3-carboxylic acid

Dissolve thiolacetic acid (0.10mL, 1.4mmol) in methanol (5mL) and treat with cesium carbonate (228mg, 0.70mmol).

20 Stir the yellow solution for 30 minutes then evaporate the solvent *in vacuo*. Dilute the resulting cesium salt with dimethylformamide (10mL) and treat with a solution of [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(R)-bromo-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]thiazepine-3-carboxylic acid (1.0mmol) in
25 tetrahydrofuran (6mL). Stir at room temperature for 2 hours, evaporate the solvent *in vacuo* and partition between ethyl acetate (75mL) and brine (50mL). Dry the organic phase (Na₂SO₄), evaporate the solvent *in vacuo* and purify by
30 chromatography to give the title compound.

Example 2

[3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-thio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-

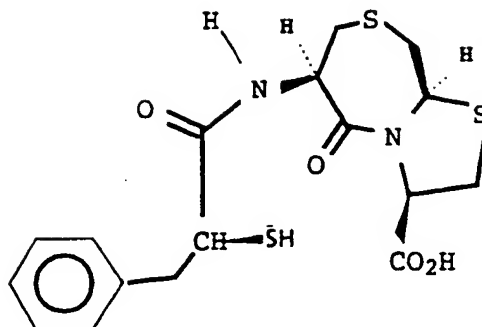
35 a][1,4]thiazepine-3-carboxylic acid

Dissolve [[3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]thiazepine-3-carboxylic acid (0.145mmol) in degassed

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5

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methanol (3mL) and tetrahydrofuran (2mL), cool in an ice bath and add lithium hydroxide (0.6mL of a 1M solution, 0.6mmol). Stir the reaction mixture for 3 hours and add 1N hydrochloric acid. Partition between methylene chloride (75mL) and water (25mL). Dry (Na₂SO₄), evaporate the solvent *in vacuo* and purify by chromatography to give the title compound.

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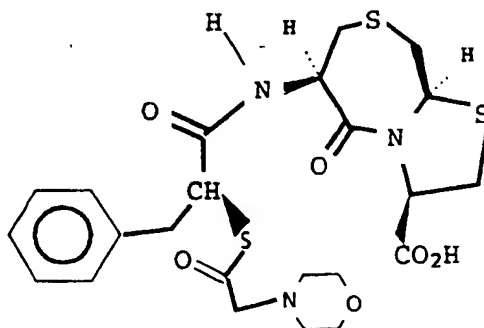
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Example 3

[3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-(4-morpholino)-
acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-
5 a][1,4]thiazepine-3-carboxylic acid



15

Suspend sodium hydride (175mg of a 60% suspension, 4.0mmol) in anhydrous dimethylformamide (4mL) and place under a nitrogen atmosphere. Bubble hydrogen sulfide gas into the suspension until solution occurs. Add triphenylmethyl 4-
20 morpholinethiolacetate (1.61g, 4.0mmol) and heat gently for 1.5 hours while bubbling nitrogen through the solution to facilitate removal of excess hydrogen sulfide gas. Add [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-oxo-2(R)-bromo-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-
25 a][1,4]thiazepine-3-carboxylic acid (2.0mmol) and stir for 2 hours. Pour into water, extract into ethyl acetate, wash with brine and dry (MgSO₄). Evaporate the solvent *in vacuo* and purify by chromatography to give the title compound.

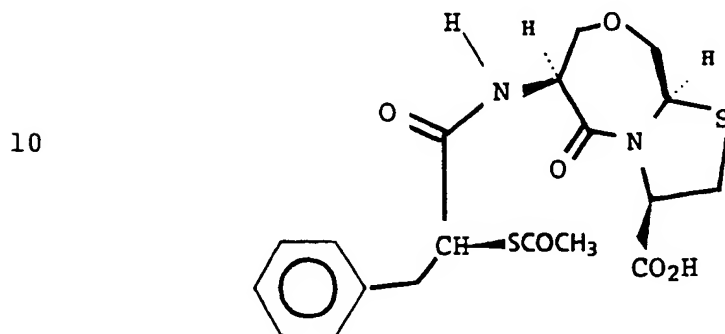
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Example 4

[3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]oxazepine-3-carboxylic acid



15

Scheme C, step a: Ethyl 2-[[[2'-carboxy-2'-benzyloxycarbonyl]ethyl]ethylether]-4(R)-thiazolidinecarboxylate

Mix L-serine methyl ester (6.6mmol) and pyridine (60mL).

20 Add, by dropwise addition, benzyl chloroformate (7.3mmol) and stir overnight. Remove excess pyridine in vacuo and dissolve the residue in a two-phase mixture of ethyl acetate/water. Separate the organic phase and extract the aqueous phase with additional ethyl acetate (2X). Wash the

25 combined organic phases with water, then brine and dry (MgSO₄). Evaporate the solvent *in vacuo* to give N-(benzyloxycarbonyl)-L-serine methyl ester.

Dissolve N-(benzyloxycarbonyl)-L-serine methyl ester

30 (63mmol) in methylene chloride/cyclohexane (1:1, 600mL). Add allyl trichloroacetimidate (26g, 128mmol) and trifluoromethanesulfonic acid (5mL, 56.6mmol). Stir at room temperature under a nitrogen atmosphere for 5 hours and dilute with methylene chloride. Wash with saturated aqueous

35 sodium hydrogen carbonate, water, dry (MgSO₄) and evaporate the solvent *in vacuo*. Purify by silica gel chromatography to give N-(benzyloxycarbonyl)-O-2-propenyl-L-serine methyl ester.

Dissolve N-(benzyloxycarbonyl)-O-2-propenyl-L-serine methyl ester (21.7mmol) in ethanol (150mL. Add 1N lithium hydroxide (50mL) and stir overnight at room temperature. Reflux for 1 hour, cool to -10°, carefully adjust to pH 4 with 1N hydrochloric acid and stir for 1 hour. Extract into ethyl acetate, dry (MgSO₄) and evaporate the solvent *in vacuo* to give N-(benzyloxycarbonyl)-O-2-propenyl-L-serine.

10

Dissolve N-(benzyloxycarbonyl)-O-2-propenyl-L-serine (29.8mmol) in methylene chloride/methanol (10:1, 220mL). Cool to -78°C and sparge with a mixture of ozone/oxygen for approximately 10 minutes until a blue color persists. Sparge with nitrogen for 10 minutes at -78°C to remove excess ozone. Treat with methylfulfide (12mL, 0.164mol) and allow to warm to room temperature. Stir at room temperature for 24 hours, evaporate the solvent *in vacuo* and dissolve the residue in ethyl acetate (200mL). Wash with water, saturated sodium chloride, dry (MgSO₄) and evaporate the solvent *in vacuo* to give N-(benzyloxycarbonyl)-O-2-oxoethyl-L-serine methyl ester.

Dissolve L-cysteine ethyl ester (12.7mmol) in tetrahydrofuran (120mL) and add N-(benzyloxycarbonyl)-O-2-oxoethyl-L-serine methyl ester (12.7mmol). Place under a nitrogen atmosphere and stir for 3 hours. Evaporate the solvent *in vacuo*, dissolve the residue in chloroform and wash with water (2X30mL). Combine the aqueous extracts and extract with chloroform (2X30mL). Combine all organic extracts, dry (Na₂SO₄) and evaporate the solvent *in vacuo* to give the title compound.

Scheme B, step b: Ethyl [3R-[3 α ,6 α , (S*),9 α]]-6-[benzyloxycarbonylamino]octahydro-5-oxothiazolo[3,2-a][1,4]oxazepine-3-carboxylate and Ethyl [3R-[3 α ,6 α , (S*),9 β]]-6-[benzyloxycarbonylamino]octahydro-5-oxothiazolo[3,2-a][1,4]oxazepine-3-carboxylate

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Dissolve ethyl 2-[[[2'-carboxy-2'-benzyloxycarbonyl]ethyl]ethylether]-4(R)-thiazolidinecarboxylate (12.7mmol) in tetrahydrofuran (30mL) and add N-ethoxycarbonyl-2-ethoxy-1,2-dihydroquinoline (EEDQ) (469mg). Stir overnight at room temperature under a nitrogen atmosphere. Evaporate the solvent *in vacuo*, partition between ethyl acetate and dilute hydrochloric acid. Separate the organic phase, wash with 5% sodium bicarbonate solution, water and brine. Dry (Na₂SO₄), evaporate the solvent *in vacuo* and purify and separate the isomers by silica gel chromatography to give the separate isomeric title compounds.

15 Scheme B, step c: [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-amino-octahydro-5-oxothiazolo[3,2-a][1,4]oxazepine-3-carboxylic acid

Dissolve ethyl [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[benzyloxycarbonylamino]octahydro-5-oxothiazolo[3,2-a][1,4]oxepine-3-carboxylate (21.7mmol) in ethanol (150mL). Add 1N lithium hydroxide (50mL) and stir overnight at room temperature. Concentrate *in vacuo*. Partition between ethyl acetate and 6N hydrochloric acid. Separate the organic phase and wash with brine. Dry (MgSO₄) and evaporate the solvent *in vacuo* to give [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[benzyloxycarbonylamino]octahydro-5-oxothiazolo[3,2-a][1,4]oxazepine-3-carboxylic acid.

Mix [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[benzyloxycarbonylamino]octahydro-5-oxothiazolo[3,2-a][1,4]oxazepine-3-carboxylic acid (27.7mmol), trifluoroacetic acid (75mL) and anisole (5mL). Stir at room temperature overnight. Pour onto water and carefully neutralize with solid sodium hydrogen carbonate. Extract into ethyl acetate (2X), wash with brine and dry (MgSO₄). Evaporate the solvent *in vacuo* and purify by chromatography to give the title compound.

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Scheme A, step a: [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(R)-bromo-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]oxazepine-3-carboxylic acid

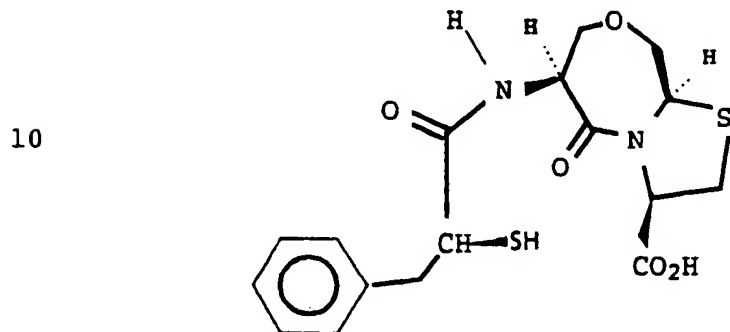
- 5 Mix 3-phenyl-2(R)-bromopropionic acid (1.0mmol) and [3R-[3 α ,6 α , (S*),9 α]]-6-amino-octahydro-5-oxothiazolo[3,2-a][1,4]oxazepine-3-carboxylic acid (1.0mmol) in methylene chloride (6mL). Add EEDQ (247mg, 1.0mmol). Stir for 15 hours at ambient temperature under argon atmosphere. Dilute
10 with ethyl acetate (25mL) and wash with 5% sulfuric acid (15mL), then saturated sodium hydrogen carbonate (15mL). Dry (Na₂SO₄), concentrate *in vacuo* and purify by silica gel chromatography to yield the title compound.

- 15 Scheme A, step b: [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]oxazepine-3-carboxylic acid

- Dissolve thiolacetic acid (0.10mL, 1.4mmol) in methanol (5mL) and treat with cesium carbonate (228mg, 0.70mmol).
20 Stir the yellow solution for 30 minutes then evaporate the solvent *in vacuo*. Dilute the resulting cesium salt with dimethylformamide (10mL) and treat with a solution of [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(R)-bromo-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]oxazepine-3-carboxylic acid (1.0mmol) in
25 tetrahydrofuran (6mL). Stir at room temperature for 2 hours, evaporate the solvent *in vacuo* and partition between ethyl acetate (75mL) and brine (50mL). Dry the organic phase (Na₂SO₄), evaporate the solvent *in vacuo* and purify by
30 chromatography to give the title compound.

Example 5

[3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-thio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]oxazepine-3-carboxylic acid



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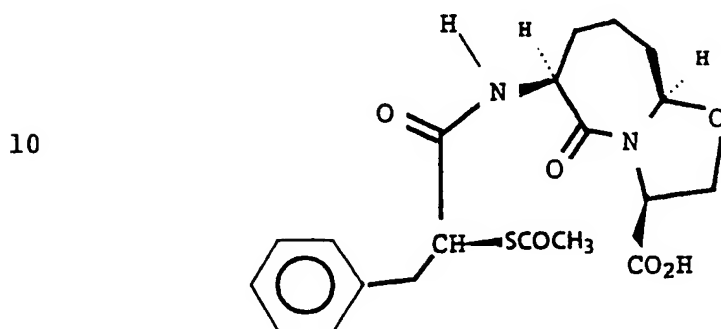
Dissolve [3R-[3 α ,6 α , (S*),9 α]]-6-[[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]oxazepine-3-carboxylic acid (0.145mmol) in degassed methanol (3mL) and tetrahydrofuran (2mL), cool in an ice bath and add lithium hydroxide (0.6mL of a 1M solution, 0.6mmol). Stir the reaction mixture for 3 hours and add 1N hydrochloric acid. Partition between methylene chloride (75mL) and water (25mL). Dry (Na₂SO₄), evaporate the solvent *in vacuo* and purify by chromatography to give the title compound.

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Example 6

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3,2-a]azepine-3-
5 carboxylic acid



Scheme B, step a: Ethyl 2-(4'-carboxy-4-phthalimidobutyl)-4(R)-oxazolidinecarboxylate

Dissolve L-serine ethyl ester (12.7mmol) in tetrahydrofuran (120mL) and add 5-formyl-2(S)-phthalimidopentanoic acid (12.7mmol). Place under a nitrogen atmosphere and stir for 3 hours. Evaporate the solvent *in vacuo*, dissolve the residue in chloroform and wash with water (2X30mL). Combine the aqueous extracts and extract with chloroform (2X30mL). Combine all organic extracts, dry (Na₂SO₄) and evaporate the solvent *in vacuo* to give the title compound.

Scheme B, step b: Ethyl [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-phthalimidooctahydro-5-oxo-oxazolo[3,2-a]azepine-3-carboxylate and Ethyl [3R-[3 α ,6 α , (S*),9 $\alpha\beta$]]-6-phthalimidooctahydro-5-oxo-oxazolo[3,2-a]azepine-3-carboxylate

Dissolve ethyl 2-(4'-carboxy-4-phthalimidobutyl)-4(R)-oxazolidinecarboxylate (12.7mmol) in tetrahydrofuran (30mL) and add N-ethoxycarbonyl-2-ethoxy-1,2-dihydroquinoline (EEDQ) (469mg). Stir overnight at room temperature under a nitrogen atmosphere. Evaporate the solvent *in vacuo*, partition between ethyl acetate and dilute hydrochloric acid. Separate the organic phase, wash with 5% sodium

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bicarbonate solution, water and brine. Dry (Na_2SO_4), evaporate the solvent *in vacuo* and purify and separate the isomers by silica gel chromatography to give the separate
5 isomeric title compounds.

Scheme B, step c: [3R-[3 α ,6 α , (S*),9 α]]-6-amino-octahydro-5-oxo-oxazolo[3,2-a]azepine-3-carboxylic acid

Dissolve ethyl [3R-[3 α ,6 α , (S*),9 α]]-6-phthalimidooctahydro-
10 5-oxo-oxazolo[3,2-a][1,4]oxepine-3-carboxylate (0.517mmol) in methanol (5mL) and treat with hydrazine monohydrate (1.1mL of a 1M solution in methanol, 1.1mmol). Stir at room temperature for 44 hours, evaporate the solvent *in vacuo* and slurry the residue in methylene chloride (10mL). Filter and
15 evaporate the solvent *in vacuo* to give ethyl [3R-[3 α ,6 α , (S*),9 α]]-6-amino-octahydro-5-oxo-oxazolo[3,2-a][1,4]oxepine-3-carboxylate.

Dissolve ethyl [3R-[3 α ,6 α , (S*),9 α]]-6-phthalimidooctahydro-
20 5-oxo-oxazolo[3,2-a]azepine-3-carboxylate (21.7mmol) in ethanol (150mL. Add 1N lithium hydroxide (50mL) and stir overnight at room temperature. Reflux for 1 hour and concentrate *in vacuo*. Partiton between ethyl acetate and 6N hydrochloric acid. Separate the organic phase and wash with
25 brine. Dry (MgSO_4) and evaporate the solvent *in vacuo* to give the title compound.

Scheme A, step a: [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(R)-bromo-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-
30 a]azepine-3-carboxylic acid

Mix 3-phenyl-2(R)-bromopropionic acid (1.0mmol) and [3R-[3 α ,6 α , (S*),9 α]]-6-amino-octahydro-5-oxo-oxazolo[3,2-a]azepine-3-carboxylic acid (1.0mmol) in methylene chloride (6mL). Add EEDQ (247mg, 1.0mmol). Stir for 15 hours at
35 ambient temperature under argon atmosphere. Dilute with ethyl acetate (25mL) and wash with 5% sulfuric acid (15mL), then saturated sodium hydrogen carbonate (15mL). Dry

(Na₂SO₄), concentrate *in vacuo* and purify by silica gel chromatography to yield the title compound.

5 Scheme A, step b: [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a]azepine-3-carboxylic acid

Dissolve thiolacetic acid (0.10mL, 1.4mmol) in methanol (5mL) and treat with cesium carbonate (228mg, 0.70mmol).

- 10 Stir the yellow solution for 30 minutes then evaporate the solvent *in vacuo*. Dilute the resulting cesium salt with dimethylformamide (10mL) and treat with a solution of [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(R)-bromo-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a]azepine-3-
- 15 carboxylic acid (1.0mmol) in tetrahydrofuran (6mL). Stir at room temperature for 2 hours, evaporate the solvent *in vacuo* and partition between ethyl acetate (75mL) and brine (50mL). Dry the organic phase (Na₂SO₄), evaporate the solvent *in vacuo* and purify by chromatography to give the title compound.

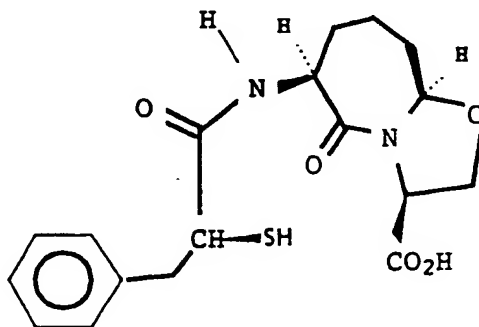
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Example 7

[3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-thio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a]azepine-3-carboxylic acid

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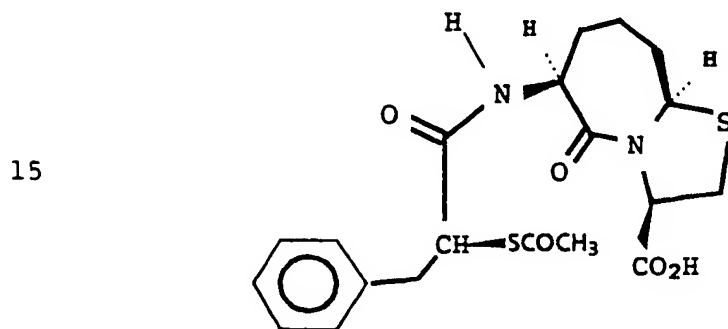
- Dissolve [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a]azepine-3-
- 35 carboxylic acid (0.145mmol) in degassed methanol (3mL) and tetrahydrofuran (2mL), cool in an ice bath and add lithium hydroxide (0.6mL of a 1M solution, 0.6mmol). Stir the

-29-

reaction mixture for 3 hours and add 1N hydrochloric acid. Partition between methylene chloride (75mL) and water (25mL). Dry (Na_2SO_4), evaporate the solvent *in vacuo* and purify by chromatography to give the title compound.

Example 8

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a]azepine-3-
carboxylic acid



Scheme A, step a: [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(R)-bromo-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a]azepine-3-carboxylic acid

Mix 3-phenyl-2(R)-bromopropionic acid (1.0mmol) and [3R-(3 α ,6 α ,9 $\alpha\beta$)-6-amino]octahydro-5-oxo-thiazolo[3,2-a]azepine-3-carboxylic acid (1.0mmol) in methylene chloride (6mL). Add EEDQ (247mg, 1.0mmol). Stir for 15 hours at ambient temperature under argon atmosphere. Dilute with ethyl acetate (25mL) and wash with 5% sulfuric acid (15mL), then saturated sodium hydrogen carbonate (15mL). Dry (Na_2SO_4), concentrate *in vacuo* and purify by silica gel chromatography to yield the title compound.

Scheme A, step b: [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a]azepine-3-carboxylic acid

Dissolve thiolacetic acid (0.10mL, 1.4mmol) in methanol (5mL) and treat with cesium carbonate (228mg, 0.70mmol). Stir the yellow solution for 30 minutes then evaporate the solvent *in vacuo*. Dilute the resulting cesium salt with

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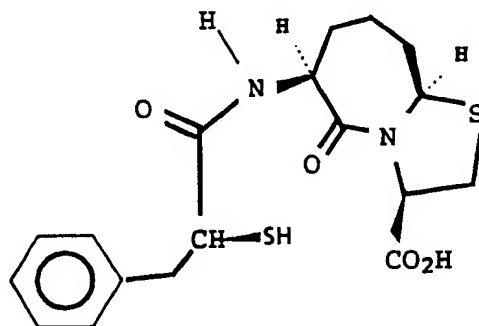
dimethylformamide (10mL) and treat with a solution of [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(R)-bromo-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a]azepine-3-carboxylic acid (1.0mmol) in tetrahydrofurn (6mL). Stir at room temperature for 2 hours, evaporate the solvent *in vacuo* and partition between ethyl acetate (75mL) and brine (50mL). Dry the organic phase (Na₂SO₄), evaporate the solvent *in vacuo* and purify by chromatography to give the title compound.

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Example 9

[3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-thio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a]azepine-3-carboxylic acid

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20

25 Dissolve [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a]azepine-3-carboxylic acid (0.145mmol) in degassed methanol (3mL) and tetrahydrofuran (2mL), cool in an ice bath, place under a nitrogen atmosphere and add lithium hydroxide (0.6mL of a 1M solution, 0.6mmol). Stir the reaction mixture for 3 hours and add 1N hydrochloric acid. Partition between methylene chloride (75mL) and water (25mL). Dry (Na₂SO₄), evaporate the solvent *in vacuo* and purify by chromatography to give the title compound.

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The following compounds can be prepared by procedures analogous to those described above in Examples 1-9:

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[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a][1,4]oxazepine-3-carboxylic acid;

5

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-(4-morpholino)acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a][1,4]oxazepine-3-carboxylic acid;

10 [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-thio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a][1,4]oxazepine-3-carboxylic acid

15 [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a][1,4]thiazepine-3-carboxylic acid;

20 [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-(4-morpholino)acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a][1,4]thiazepine-3-carboxylic acid;

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-thio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]thiazepine-3-carboxylic acid;

25

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-(4-morpholino)acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]oxazepine-3-carboxylic acid;

30 [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-(4-morpholino)acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a]azepine-3-carboxylic acid; and

35 [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-(4-morpholino)acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a]azepine-3-carboxylic acid.

As used herein, the term "patient" refers to warm-blooded animals or mammals, including mice, rats and humans. A patient is in need of treatment to inhibit enkephalinase 5 when the patient is suffering from acute or chronic pain and is in need of an endorphin- or enkephalin-mediated analgesic effect. In addition, a patient is in need of treatment to inhibit enkephalinase when the patient is suffering from a disease state characterized by abnormalities in fluid, 10 electrolyte, blood pressure, intraocular pressure, renin, or aldosterone homeostasis, such as, but not limited to, hypertension, renal diseases, hyperaldosteronemia, cardiac hypertrophy, glaucoma and congestive heart failure. In these instances the patient is in need of an ANP-mediated 15 diuretic, natriuretic, hypotensive, hypoaldosteronemic effect. Inhibition of enkephalinase would provide an endorphin- or enkephalin-mediated analgesic effect by inhibiting the metabolic degradation of endorphins and enkephalins. Inhibition of enkephalinase would provide an 20 ANP-mediated diuretic, natriuretic, hypotensive, hypoaldosteronemic effect by inhibiting the metabolic degradation of ANP. Inhibition of enkephalinase would also potentiate endogenous levels of bradykinin. Inhibition of enkephalinase would also modulate intestinal smooth muscle 25 contractility and would be useful in the treatment of irritable bowel syndrome.

In addition, a patient is in need of treatment to inhibit enkephalinase when the patient is in need of an 30 antidepressant effect or a reduction in severity of withdrawal symptoms associated with termination of opiate or morphine administration.

The identification of those patients who are in need of 35 treatment to inhibit enkephalinase is well within the ability and knowledge of one skilled in the art. A clinician skilled in the art can readily identify, by the use of clinical tests, physical examination and

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medical/family history, those patients who are in need of an endorphin- or enkephalin-mediated analgesic effect or who are in need of an ANP-mediated diuretic, natriuretic, 5 hypotensive or hypoaldosteronemic effect.

An effective enkephalinase inhibitory amount of a compound of Formula (I) is an amount which is effective in inhibiting enkephalinase and in thus inhibiting the 10 metabolic degradation of the naturally-occurring circulating regulatory peptides such as the endorphins, including enkephalins, and ANP. Successful treatment is also understood to include prophylaxis in treating a patient in those instances such as, for example, in a pre-operative 15 procedure, where a patient will be suffering from acute or chronic pain in the near future.

An effective enkephalinase inhibitory amount of a compound of Formula (I) is an amount which is effective in 20 inhibiting enkephalinase in a patient in need thereof which results, for example, in endorphin- or enkephalin-mediated analgesic effects or in ANP-mediated diuretic, natriuretic, hypotensive, hypoaldosteronemic effect.

25 An effective enkephalinase inhibitory dose can be readily determined by the use of conventional techniques and by observing results obtained under analogous circumstances. In determining the effective dose, a number of factors are considered including, but not limited to: the species of 30 patient; its size, age, and general health; the specific disease involved; the degree of or involvement or the severity of the disease; the response of the individual patient; the particular compound administered; the mode of administration; the bioavailability characteristics of the 35 preparation administered; the dose regimen selected; and the use of concomitant medication.

An effective enkephalinase inhibitory amount of a compound of Formula (I) will generally vary from about 0.01 milligram per kilogram of body weight per day (mg/kg/day) to about 20 mg/kg/day. A daily dose of from about 0.1 mg/kg to about 10 mg/kg is preferred.

In addition, the present invention further provides a method of inhibiting ACE in a patient in need thereof comprising administering to said patient an effective ACE inhibitory amount of a compound of Formula (I). A patient is in need of treatment to inhibit ACE when the patient is suffering from hypertension, chronic congestive heart failure, hyperaldosteronemia or cognitive disorders.

15 Inhibition of ACE reduces levels of angiotensin II and thus inhibits the vasopressor, hypertensive and hyperaldosteronemic effects caused thereby. An effective ACE inhibitory amount of a compound of Formula (I) is that amount which is effective in inhibiting ACE in a patient in need thereof which results, for example, in a hypotensive effect. An effective ACE inhibitory amount and an effective ACE inhibitory dose are the same as that described above for an effective enkephalinase inhibitory amount and dose.

25 In addition, the present invention further provides a method for treating a patient suffering from smooth cell proliferation. An effective smooth cell proliferation inhibitory amount of a compound of Formula (I) is that amount which is effective in inhibiting smooth cell proliferation in a patient in need thereof which results, for example, in a reduced myointimal thickening after vascular injury. An effective smooth cell proliferation inhibitory amount and an effective smooth cell proliferation inhibitory dose are the same as that described above for an effective enkephalinase inhibitory amount and dose.

30 proliferation in a patient in need thereof which results, for example, in a reduced myointimal thickening after vascular injury. An effective smooth cell proliferation inhibitory amount and an effective smooth cell proliferation inhibitory dose are the same as that described above for an

35 effective enkephalinase inhibitory amount and dose.

In effecting treatment of a patient, compounds of Formula (I) can be administered in any form or mode which

-35-

makes the compound bioavailable in effective amounts, including oral and parenteral routes. For example, the compound can be administered orally, subcutaneously, 5 intramuscularly, intravenously, transdermally, intranasally, rectally, and the like. Oral administration is generally preferred. One skilled in the art of preparing Formulations can readily select the proper form and mode of administration depending upon the disease state to be 10 treated, the stage of the disease, and other relevant circumstances.

Compounds of Formula (I) can be administered in the form of pharmaceutical compositions or medicaments which are made 15 by combining the compounds of Formula (I) with pharmaceutically acceptable carriers or excipients, the proportion and nature of which are determined by the chosen route of administration, and standard pharmaceutical practice.

20

In another embodiment, the present invention provides compositions comprising a compound of Formula (I) in admixture or otherwise in association with one or more inert carriers. These compositions are useful, for 25 example, as assay standards, as convenient means of making bulk shipments, or as pharmaceutical compositions. An assayable amount of a compound of Formula (I) is an amount which is readily measurable by standard assay procedures and techniques as are well known and appreciated by those 30 skilled in the art. Assayable amounts of a compound of Formula (I) will generally vary from about 0.001% to about 75% of the composition by weight. Inert carriers can be any material which does not degrade or otherwise covalently react with a compound of Formula (I). Examples of suitable 35 inert carriers are water; aqueous buffers, such as those which are generally useful in High Performance Liquid Chromatography (HPLC) analysis; organic solvents, such as

acetonitrile, ethyl acetate, hexane and the like; and pharmaceutically acceptable carriers or excipients.

5 More particularly, the present invention provides pharmaceutical compositions comprising an effective amount of a compound of Formula (I) in admixture or otherwise in association with one or more pharmaceutically acceptable carriers or excipients.

10

The pharmaceutical compositions or medicaments are prepared in a manner well known in the pharmaceutical art. The carrier or excipient may be a solid, semi-solid, or liquid material which can serve as a vehicle or medium for
15 the active ingredient. Suitable carriers or excipients are well known in the art. The pharmaceutical composition may be adapted for oral or parenteral use and may be administered to the patient in the form of tablets, capsules, suppositories, solution, suspensions, or the like.

20

The pharmaceutical compositions may be administered orally, for example, with an inert diluent or with an edible carrier. They may be enclosed in gelatin capsules or compressed into tablets. For the purpose of oral
25 therapeutic administration, the compounds of Formula (I) may be incorporated with excipients and used in the form of tablets, troches, capsules, elixirs, suspensions, syrups, wafers, chewing gums and the like. These preparations should contain at least 4% of the compound of Formula (I),
30 the active ingredient, but may be varied depending upon the particular form and may conveniently be between 4% to about 70% of the weight of the unit. The amount of the active ingredient present in compositions is such that a unit dosage form suitable for administration will be obtained.

35

The tablets, pills, capsules, troches and the like may also contain one or more of the following adjuvants: binders, such as microcrystalline cellulose, gum tragacanth

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or gelatin; excipients, such as starch or lactose, disintegrating agents such as alginic acid, Primogel, corn starch and the like; lubricants, such as magnesium stearate
5 or Sterotex; glidants, such as colloidal silicon dioxide; and sweetening agents, such as sucrose or saccharin may be added or flavoring agents, such as peppermint, methyl salicylate or orange flavoring. When the dosage unit form is a capsule, it may contain, in addition to materials of
10 the above type, a liquid carrier such as polyethylene glycol or a fatty oil. Other dosage unit forms may contain other various materials which modify the physical form of the dosage unit, for example, as coatings. Thus, tablets or pills may be coated with sugar, shellac, or other enteric
15 coating agents. A syrup may contain, in addition to the active ingredient, sucrose as a sweetening agent and certain preservatives, dyes and colorings and flavors. Materials used in preparing these various compositions should be pharmaceutically pure and non-toxic in the amounts used.

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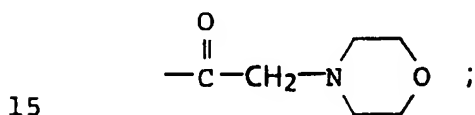
For the purpose of parenteral administration, the compounds of Formula (I) may be incorporated into a solution or suspension. These preparations should contain at least 0.1% of a compound of the invention, but may be varied to be
25 between 0.1 and about 50% of the weight thereof. The amount of the active ingredient present in such compositions is such that a suitable dosage will be obtained.

The solutions or suspensions may also include one or
30 more of the following adjuvants: sterile diluents such as water for injection, saline solution, fixed oils, polyethylene glycols, glycerine, propylene glycol or other synthetic solvents; antibacterial agents such as benzyl alcohol or methyl paraben; antioxidants such as ascorbic
35 acid or sodium bisulfite; chelating agents such as ethylene diaminetetraacetic acid; buffers such as acetates, citrates or phosphates and agents for the adjustment of toxicity such as sodium chloride or dextrose. The parenteral preparation

can be enclosed in ampules, disposable syringes or multiple dose vials made of glass or plastic.

5 As with any group of structurally related compounds which possess a particular generic utility, certain groups and configurations are preferred for compounds of Formula (I) in their end-use application.

10 The compounds of Formula (I) wherein R₁ is acetyl or a group of the formula



R₂ is an Ar-Y group wherein Ar is phenyl, 4,5-methylenedioxyphenyl or 2-benzofuranyl and Y is -CH₂-; A is -S- or -CH₂-; B is -S- and R is hydrogen, ethyl or benzyl
20 are preferred.

It is, of course, understood that the compounds of Formula (I) may exist in a variety of isomeric configurations including structural as well as stereo
25 isomers. It is further understood that the present invention encompasses those compounds of Formula (I) in each of their various structural and stereo isomeric configurations as individual isomers and as mixtures of isomers.

30

The following specific compounds of Formula (I) are particularly preferred in the end-use application of the compounds of the present invention:

35 [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]thiazepine-3-carboxylic acid;

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-thio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]thiazepine-3-carboxylic acid;

5

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-(4-morpholino)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]thiazepine-3-carboxylic acid;

10 [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]oxazepine-3-carboxylic acid;

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-thio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]oxazepine-3-carboxylic acid;

15

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a]azepine-3-carboxylic acid;

20

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-thio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a]azepine-3-carboxylic acid;

25

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a]azepine-3-carboxylic acid;

30 [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-thio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a]azepine-3-carboxylic acid;

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a][1,4]oxazepine-3-carboxylic acid;

35

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-thio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a][1,4]oxazepine-3-carboxylic acid

5

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a][1,4]thiazepine-3-carboxylic acid;

10 [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-(4-morpholino)acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a][1,4]thiazepine-3-carboxylic acid;

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-thio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]thiazepine-3-carboxylic acid;

15

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-(4-morpholino)acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]oxazepine-3-carboxylic acid;

20

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-(4-morpholino)acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a][1,4]oxazepine-3-carboxylic acid;

25

[3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-(4-morpholino)acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a]azepine-3-carboxylic acid; and

30 [3R-[3 α ,6 α , (S*),9 $\alpha\alpha$]]-6-[[1-Oxo-2(S)-(4-morpholino)acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a]azepine-3-carboxylic acid.

The following studies illustrate the utility of the compounds of the present invention as enkephalinase inhibitors and as ACE inhibitors.

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Enkephalinase is partially purified from rat kidney. The enzyme is extracted from the microvilli fraction by using Triton X-100 according to the method of Malfroy and Schwartz [*J. Biol. Chem.* 259, 14365-14370 (1984)] or by using a proteolytic treatment according to the method of Almenoff and Orlowski [*Biochem.* 22, 590-599 (1983)]. The enzyme is further purified by anion exchange chromatography (Mono Q[™] column, Pharmacia) using a Pharmacia FPLC system. The enzyme activity may be measured by the fluorometric methods of Florentin et al. [*Anal. Biochem.* 141, 62-69 (1984)] or of Almenoff and Orlowski [*J. Neurochemistry* 42, 151-157 (1984)]. The enzyme is assayed in 50mM HEPES buffer (pH 7.4) in a 3.0 mL reaction volume containing 12 μ M of the substrate dansyl-D-AlaGly(p-nitro)PheGly ($K_m=40\mu$ M) at 25°C. The substrate (and inhibitor) is added from a concentrated stock solution in DMSO (up to 0.1 mL DMSO final volume). The enzyme in a small volume (approximately 0.1 μ g of FPLC purified protein) is added to initiate the reaction and the rate of fluorescence increase is recorded continuously using a fluorometer (excitation at 339nm, emission at 562nm).

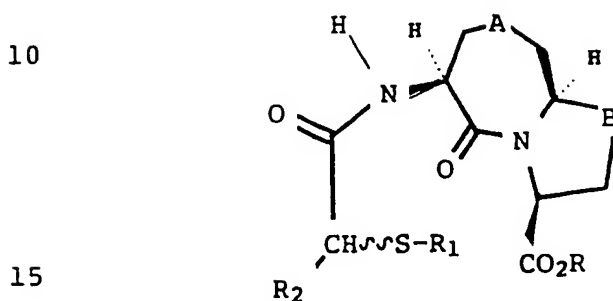
The enzymatic activity of ACE is monitored using the spectrophotometric substrate described by Holmquist et al. [*Anal. Biochem.* 95, 540-548 (1979)] and the buffer system described by Ryan [*Methods of Enzymatic Analysis*, 3rd ed., H. U. Bergmeyer, editor; vol. V, Verlag Chemie, Weinheim, 1983, pp. 20-34].

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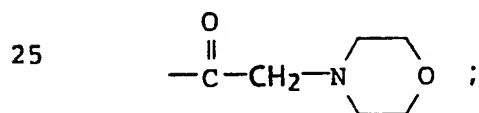
WHAT IS CLAIMED IS:

- 5 1. A compound of the formula



wherein

- 20 R is hydrogen, a C₁-C₄ alkyl or an Ar-Y- group,
 -CH₂O-C(O)C(CH₃)₃ or diphenylmethyl;
 R₁ is hydrogen, acetyl, -CH₂O-C(O)C(CH₃)₃ or benzoyl or a
 group of the formula



- 30 R₂ is hydrogen, C₁-C₈ alkyl, -CH₂OCH₂CH₂OCH₃ or an Ar-Y-
 group;
 A is -CH₂-, -O-, or -S-;
 B is -S- or -O-; and
 the pharmaceutically acceptable salts thereof.

2. A compound according to Claim 1 wherein A is -S-.

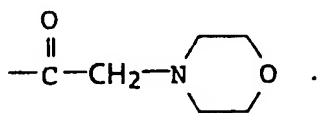
5 3. A compound according to Claim 2 wherein B is -S-.

4. A compound according to Claim 3 wherein R₂ is phenylmethyl.

10 5. A compound according to Claim 4 wherein R₁ is acetyl.

6. A compound according to Claim 4 wherein R₁ is a group of the formula

15



20 7. A compound according to Claim 1 wherein A is -CH₂-.

8. A compound according to Claim 7 wherein B is -S-.

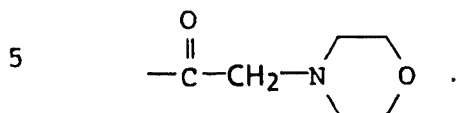
9. A compound according to Claim 8 wherein R₂ is
25 phenylmethyl.

10. A compound according to Claim 9 wherein R₁ is acetyl.

30

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11. A compound according to Claim 10 wherein R₁ is a group of the formula



12. A compound of Claim 1 wherein the compound is [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]thiazepine-3-carboxylic acid.

13. A compound of Claim 1 wherein the compound is [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-(4-morpholino)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a][1,4]thiazepine-3-carboxylic acid.

14. A compound of Claim 1 wherein the compound is [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a]azepine-3-carboxylic acid.

15. A compound of Claim 1 wherein the compound is [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a]azepine-3-carboxylic acid.

16. A compound of Claim 1 wherein the compound is [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a][1,4]thiazepine-3-carboxylic acid.

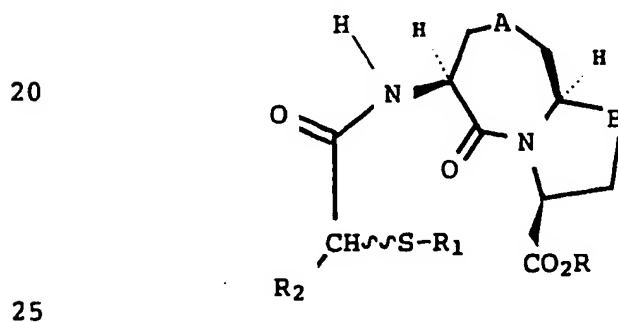
17. A compound of Claim 1 wherein the compound is [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-(4-morpholino)acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a][1,4]thiazepine-3-carboxylic acid.

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18. A compound of Claim 1 wherein the compound is [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-(4-morpholino)acetylthio-3-phenylpropyl]amino]octahydro-5-oxo-oxazolo[3-2-a]azepine-3-
5 carboxylic acid.

19. A compound of Claim 1 wherein the compound is [3R-[3 α ,6 α , (S*),9 α]]-6-[[1-Oxo-2(S)-(4-morpholino)acetylthio-3-phenylpropyl]amino]octahydro-5-oxothiazolo[3-2-a]azepine-3-
10 carboxylic acid.

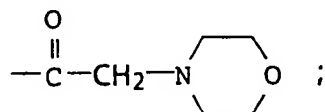
20. A method of inhibiting enkephalinase in a patient in need thereof comprising administering to said patient an effective enkephalinase inhibitory amount of a compound of
15 the formula



wherein

R is hydrogen, a C₁-C₄ alkyl or an Ar-Y- group,
-CH₂O-C(O)C(CH₃)₃ or diphenylmethyl;

30 R₁ is hydrogen, acetyl, -CH₂O-C(O)C(CH₃)₃ or benzoyl or a group of the formula



R_2 is hydrogen, C_1 - C_8 alkyl, $-\text{CH}_2\text{OCH}_2\text{CH}_2\text{OCH}_3$ or an Ar-Y-group;

A is $-\text{CH}_2-$, $-\text{O}-$, or $-\text{S}-$;

5 B is $-\text{S}-$ or $-\text{O}-$; and

the pharmaceutically acceptable salts thereof.

21. A method according to Claim 20 wherein the patient
is in need of an endorphin- or enkephalin-mediated analgesic
10 effect.

22. A method according to Claim 20 wherein the patient
is in need of an ANP-mediated hypotensive effect.

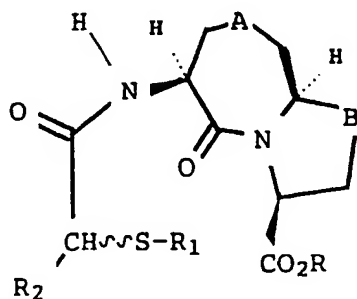
15 23. A method according to Claim 20 wherein the patient
is in need of an ANP-mediated diuretic effect.

24. A method according to Claim 20 wherein the patient
is suffering from congestive heart failure.
20

25. A method according to Claim 20 wherein the patient
is suffering from irritable bowel syndrome.

26. A method of inhibiting ACE in a patient in need
25 thereof comprising administering to said patient an
effective ACE inhibitory amount of a compound of the formula

30

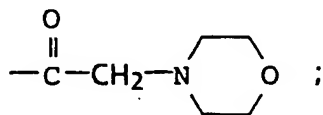


35

wherein

R is hydrogen, a C₁-C₄ alkyl or an Ar-Y- group,
-CH₂O-C(O)C(CH₃)₃ or diphenylmethyl;

5 R₁ is hydrogen, acetyl, -CH₂O-C(O)C(CH₃)₃ or benzoyl or a
group of the formula



10 R₂ is hydrogen, C₁-C₈ alkyl, -CH₂OCH₂CH₂OCH₃ or an Ar-Y-
group;

A is -CH₂-, -O-, or -S-;

B is -S- or -O-; and

15 the pharmaceutically acceptable salts thereof.

27. A method according to Claim 26 wherein the patient
is in need of a hypotensive effect.

20 28. A method according to Claim 26 wherein the patient
is in need of a cognition enhancing effect.

29. A method according to Claim 26 wherein the patient
is suffering from congestive heart failure.

25 30. A method of inhibiting smooth cell proliferation
in a patient in need thereof comprising administering to
said patient an effective smooth cell proliferation
inhibitory amount of a compound of the formula
wherein

30 R is hydrogen, a C₁-C₄ alkyl or an Ar-Y- group,
-CH₂O-C(O)C(CH₃)₃ or diphenylmethyl;

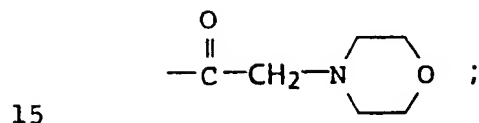
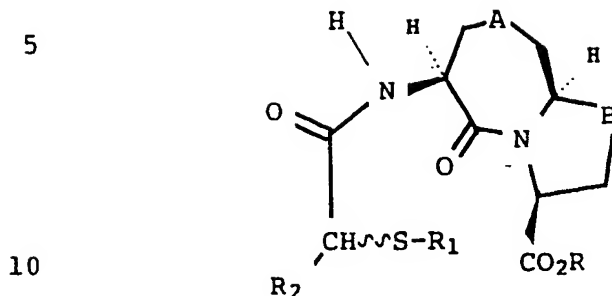
R₁ is hydrogen, acetyl, -CH₂O-C(O)C(CH₃)₃ or benzoyl or a
group of the formula

35 R₂ is hydrogen, C₁-C₈ alkyl, -CH₂OCH₂CH₂OCH₃ or an Ar-Y-
group;

A is -CH₂-, -O-, or -S-;

B is -S- or -O-; and

the pharmaceutically acceptable salts thereof.



31. A composition comprising an assayable amount of a compound of Claim 1 in admixture or otherwise in association with an inert carrier.

32. A pharmaceutical composition comprising an effective immunosuppressive amount of a compound of Claim 1 in admixture or otherwise in association with one or more pharmaceutically acceptable carriers or excipients.

33. A compound according to Claim 1 for use as a pharmaceutically active compound.

34. A compound according to any one of Claims 1-19 for the inhibition of enkephalinase.

35. A compound according to any one of Claims 1-19 for use in the treatment of acute or chronic pain.

36. A compound according to any one of Claims 1-19 for use as an antihypotensive agent in the treatment of congestive heart failure.

37. A compound according to any one of Claims 1-19 for use as an antihypotensive agent in the treatment of cardiac hypertrophy.

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38. A compound according to any one of Claims 1-19 for use in the treatment of congestive heart failure.

39. A compound according to any one of Claims 1-19 for
10 use in the treatment of cardiac hypertrophy.

40. A compound according to any one of Claims 1-19 for use in the treatment of irritable bowel syndrome.

15 41. A compound according to any one of Claims 1-19 for use as a diuretic.

42. A compound according to any one of Claims 1-19 for the inhibition of ACE.

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43. A compound according to any one of Claims 1-19 for the treatment of loss of cognitive function.

44. A compound according to any one of Claims 1-19 for
25 the inhibition of smooth cell proliferation.

45. The use of a compound according to any one of Claims 1-19, optionally in combination with a pharmaceutically acceptable carrier, for the preparation of
30 a pharmaceutical composition for the treatment of hypertension, acute or chronic pain, congestive heart failure, cardiac hypertrophy, irritable bowel syndrome, loss of cognitive function or as a diuretic.

35 46. The use of a compound according to any one of Claims 1-19, optionally in combination with a pharmaceutically acceptable carrier, for the preparation of an enkephalinase inhibitor.

47. The use of a compound according to any one of
Claims 1-19, optionally in combination with a
5 pharmaceutically acceptable carrier, for the preparation of
an ACE inhibitor.

48. The use of a compound according to any one of
Claims 1-19, optionally in combination with a
10 pharmaceutically acceptable carrier, for the preparation of
an smooth cell proliferation inhibitor.

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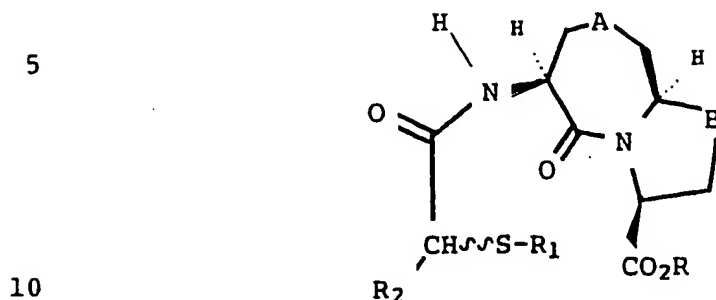
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-51-

49. A process for preparing a compound of the formula



wherein

R is hydrogen;

R₁ is acetyl or benzoyl;

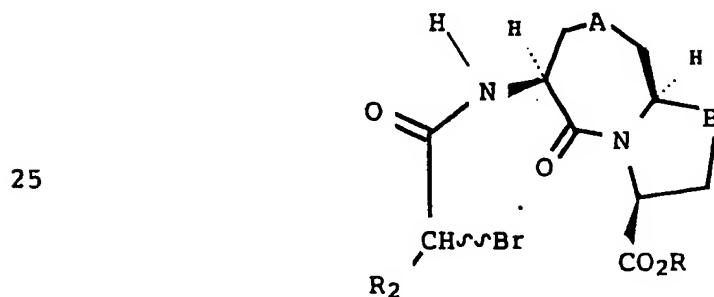
15 R₂ is hydrogen, C₁-C₈ alkyl, -CH₂OCH₂CH₂OCH₃ or an Ar-Y-group;

A is -CH₂-, -O-, or -S-; and

B is -S- or -O-;

comprising reacting a compound of the formula

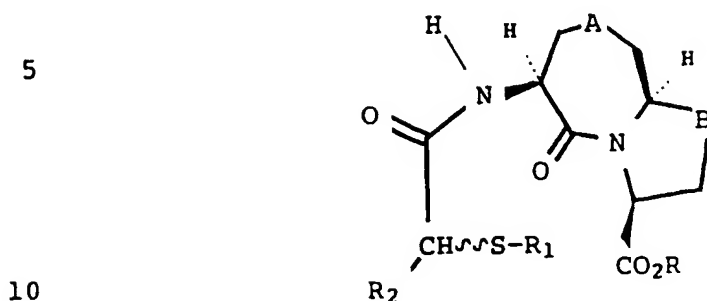
20



wherein R, R₂, A and B are defined above with a compound of the formula R₁SH, wherein R₁ is defined above in the presence of an appropriate base.

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50. A process for preparing a compound of the formula



wherein

R is hydrogen;

R₁ is hydrogen

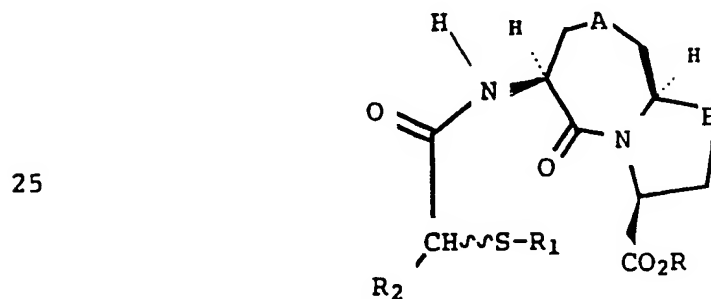
15 R₂ is hydrogen, C₁-C₈ alkyl, -CH₂OCH₂CH₂OCH₃ or an Ar-Y-group;

A is -CH₂-, -O-, or -S-; and

B is -S- or -O-,

comprising reacting a compound of the formula

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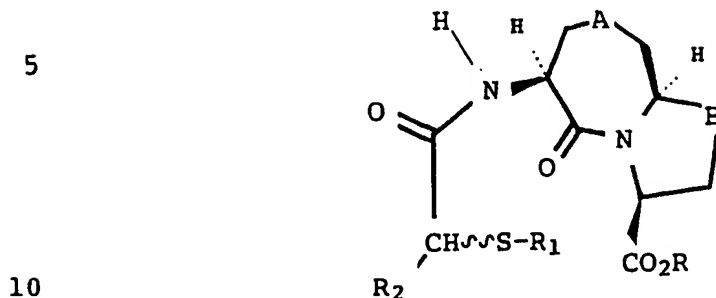


wherein R, R₂, A and B are defined above and R₁ is acetyl or benzoyl with lithium hydroxide in a suitable solvent.

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-53-

51. A process for preparing a compound of the formula



wherein

R is a C₁-C₄ alkyl or an Ar-Y- group,
-CH₂O-C(O)C(CH₃)₃ or diphenylmethyl;

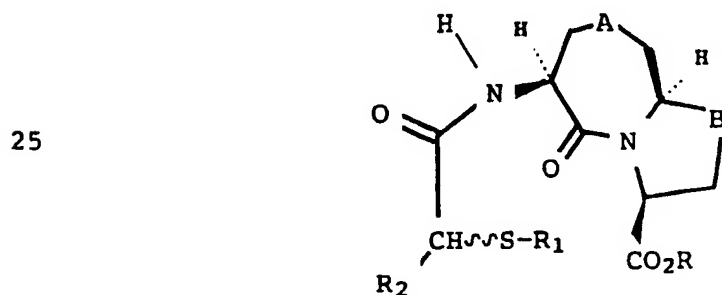
15 R₁ is acetyl or benzoyl;

R₂ is hydrogen, C₁-C₈ alkyl, -CH₂OCH₂CH₂OCH₃ or an Ar-Y-
group;

A is -CH₂-, -O-, or -S-; and

B is -S- or -O-;

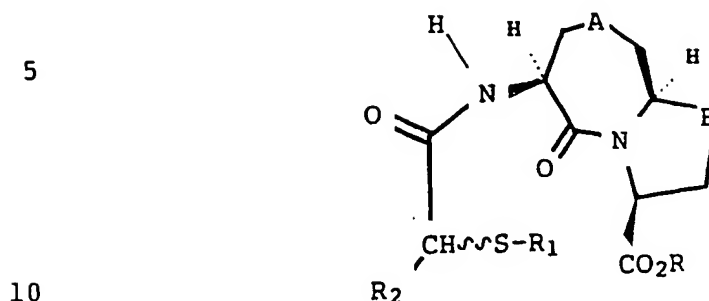
20 comprising reacting a compound of the formula



wherein R₁, R₂, A, B are defined above and R is hydrogen with
an appropriate alkyl halide in the presence of a non-
nucleophilic base in a suitable aprotic solvent.

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52. A process for preparing a compound of the formula



wherein

R is hydrogen, a C₁-C₄ alkyl or an Ar-Y- group,
-CH₂O-C(O)C(CH₃)₃ or diphenylmethyl;

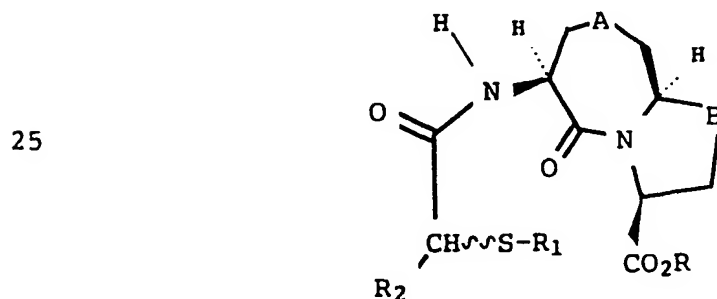
15 R₁ is hydrogen

R₂ is hydrogen, C₁-C₈ alkyl, -CH₂OCH₂CH₂OCH₃ or an Ar-Y-
group;

A is -CH₂-, -O-, or -S-; and

B is -S- or -O-;

20 comprising reacting a compound of the formula

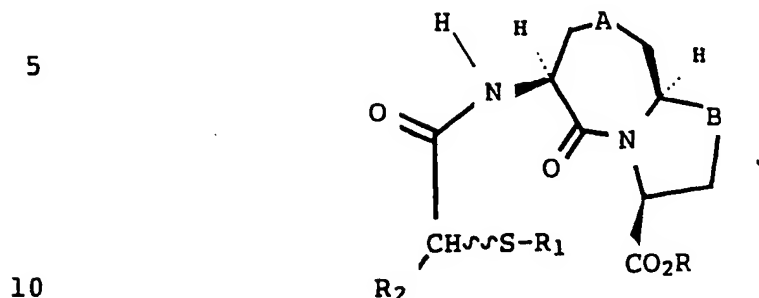


30 wherein R, R₂, A and B are defined above and R₁ is acetyl or
benzoyl with ammonia in a suitable protic solvent.

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-55-

53. A process for preparing a compound of the formula



wherein

R is hydrogen, C₁-C₄ alkyl or an Ar-Y- group,
-CH₂O-C(O)C(CH₃)₃ or diphenylmethyl;

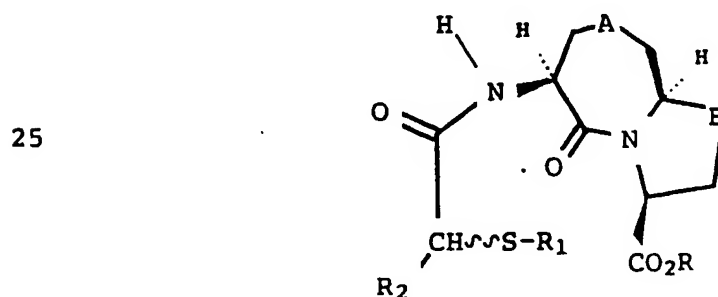
15 R₁ is -CH₂O-C(O)C(CH₃)₃;

R₂ is hydrogen, C₁-C₈ alkyl, -CH₂OCH₂CH₂OCH₃ or an Ar-Y-
group;

A is -CH₂-, -O-, or -S-; and

B is -S- or -O-;

20 comprising reacting a compound of the formula

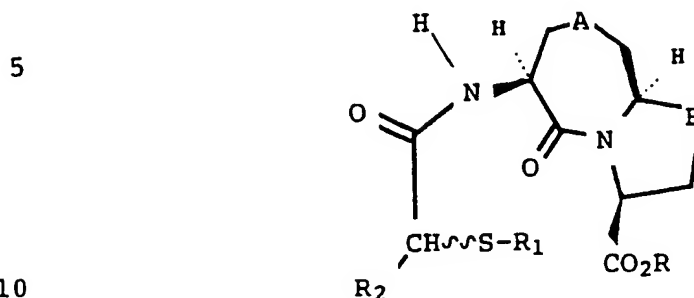


30 wherein R, R₂, A and B are defined above and R₁ is hydrogen
with chloromethyl pivalate in a suitable aprotic solvent in
the presence of a non-nucleophilic base.

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-56-

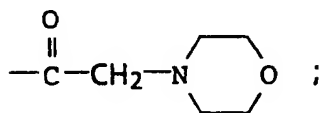
54. A process for preparing a compound of the formula



wherein

R is hydrogen, a C₁-C₄ alkyl or an Ar-Y- group,
-CH₂O-C(O)C(CH₃)₃ or diphenylmethyl;

15 R₁ is a group of the formula

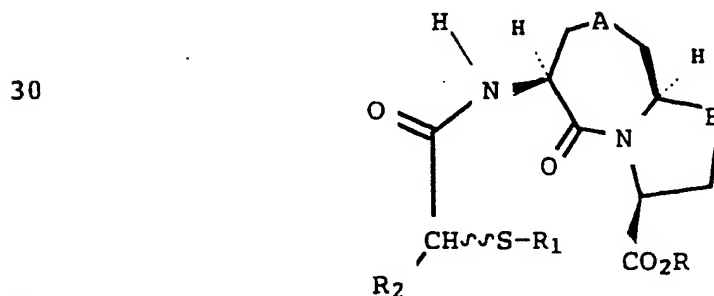


R₂ is hydrogen, C₁-C₈ alkyl, -CH₂OCH₂CH₂OCH₃ or an Ar-Y-
group;

A is -CH₂-, -O-, or -S-;

B is -S- or -O-; and

25 the pharmaceutically acceptable salts thereof
comprising reacting a compound of the formula



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wherein R, R₂, A and B are defined as above and R₁ is H with 4-morpholinethiolacetate in the presence of a suitable coupling agent in a suitable aprotic solvent.

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INTERNATIONAL SEARCH REPORT

International Application No.
PCT/US 93/09001

A. CLASSIFICATION OF SUBJECT MATTER
IPC 5 C07K5/06 A61K37/64

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 5 C07K A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	EP,A,0 481 522 (MERRELL DOW PHARMACEUTICALS INC.) 22 April 1992 see the whole document ---	1-54
A	BIOCHEMICAL AND BIOPHYSICAL RESEARCH COMMUNICATIONS. vol. 117, no. 1, 30 November 1983, DULUTH, MINNESOTA US pages 108 - 113 W.H.PARSONS 'BENZOLACTAMS.A NEW CLASS OF CONVERTING ENZYME INHIBITORS' The whole document ;see especially Table 1 ---	1-54
A	EP,A,0 240 366 (SANKYO COMPANY LTD) 7 October 1987 The whole document;see especially Table 6 -----	1-54

☐ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

* Special categories of cited documents :

- *A* document defining the general state of the art which is not considered to be of particular relevance
- *E* earlier document but published on or after the international filing date
- *L* document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- *O* document referring to an oral disclosure, use, exhibition or other means
- *P* document published prior to the international filing date but later than the priority date claimed

- *T* later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
- *X* document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
- *Y* document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.
- *&* document member of the same patent family

Date of the actual completion of the international search

15 December 1993

Date of mailing of the international search report

18 -01- 1994

Name and mailing address of the ISA
European Patent Office, P.B. 5818 Patentlaan 2
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Authorized officer

Groenendijk, M

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 93/09001

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:
Remark : Although claims 20-30 are directed to a method of treatment of (diagnostic method practised on) the human/animal body the search has been carried out and based on the alleged effects of the compound/ composition.
2. ☐ Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

Information on patent family members

al Application No

/US 93/09001

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
EP-A-0481522	22-04-92	AU-B- 642535	21-10-93
		AU-A- 8581991	30-04-92
		CN-A- 1061971	17-06-92
		JP-A- 4282382	07-10-92

EP-A-0240366	07-10-87	AU-B- 599642	26-07-90
		AU-A- 7111587	08-10-87
		JP-A- 63045262	26-02-88
		SU-A- 1598876	07-10-90
		US-A- 4778790	18-10-88
		JP-A- 63152378	24-06-88
